A Technique for Rearing the Immature Stages of Tabanidae (Diptera)[1]

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Most species of Tabanidae for which information about the larval stage is available have been found in semi-aquatic environments, and one of the principal problems in rearing the immature stages has been how to simulate such an environment.

Two rearing methods have been employed. Hine (1906) used jelly glasses that contained sand covered with algae or leaves of water plants, while others have modified this method by using material from larval habitats. Marchand (1917) reared larvae in 7-in. test tubes fitted with 6-in. rolls of filter paper kept moist by a small amount of water in the bottoms of the tubes. This method was better since it allowed the larvae to be observed. However, the tubes needed daily attention to prevent dessication. Attempts to reduce evaporation by corking the containers might have caused the larvae to be asphyxiated, especially in smaller vessels.

Roberts and Dicke (1964) described the use of plastic containers lined with filter paper. They noted a positive thigmotactic response of the larvae, i.e., larvae invariably were located between the container wall and the filter paper. In addition, they noted in large larvae, 10 to 15 mm or longer, a greater mortality because of incomplete ecdysis or a sealing of the anus by crusted fecal material. These difficulties indicated that, in nature, both ecdysis and evacuation of the gut are assisted by the mechanical resistance of the medium to the movement of the larvae.

Thus, a medium had to be developed in which (1) the larvae could easily be observed, (2) a wet environment could be maintained without constant attention, and (3) there was enough mechanical resistance to aid molting and evacuation of the gut.

[1] In cooperation with the Delta Branch of the Mississippi Agricultural Experiment Station.

Rearing Technique.—Two materials were used successfully in rearing various species of Tabanidae. The first consisted of glass beads 4, 5, or 6 mm in diameter barely covered with water. These were satisfactory for larvae 10 mm long or longer but not for early instars, since these smaller larvae and their exuviae were extremely difficult to locate in the beads.

The second material was agar. Nutrient bacterial agar, as a medium and food source, proved unsatisfactory because of the rapid growth of bacteria. However, agar, which by itself neither supports bacteria nor serves as a food source, was used successfully as a larval medium. The most suitable concentrations ranged from 0.8 to 1%. At less than 0.8% the medium was too soft to provide sufficient resistance, and above 1% it was too hard. Newly hatched and other small larvae had difficulty in penetrating the harder medium, whereas large larvae broke it apart, which seriously interfered with visual observation. In the 0.8–1% range, however, the small larvae were able to penetrate the medium and move easily about in it. With the larger larvae the medium closed around them and prevented visual distortion.

Even in plain agar waste materials and excess food supported bacterial growth, and in order to use the medium for longer periods, an antibiotic, Panalba®,[2] was added. This contains two parts tetracycline phosphate and one part novobiocin. The finished agar had 25 μg/ml of tetracycline phosphate and 12.5 μg/ml of novobiocin, and could be used 7–10 days, whereas untreated agar could be used only 3–5 days before contamination necessitated transferring the larvae to fresh medium. Recently, a second antibiotic, Pimafucin®,[2] used in conjunction with Panalba at a concentration of 30–40 μg/ml has extended the life of the medium to 2–3 weeks.

The present technique consists in rearing the larvae in the agar medium until they are 15–20 mm long. Then they are transferred to the glass bead substrate. The pupae are em-

[2] Mention of proprietary products does not necessarily imply their endorsement by the USDA.

bedded in an agar medium in an upright position, with the thoracic spiracles protruding above the surface, a position that appears to facilitate eclosion.

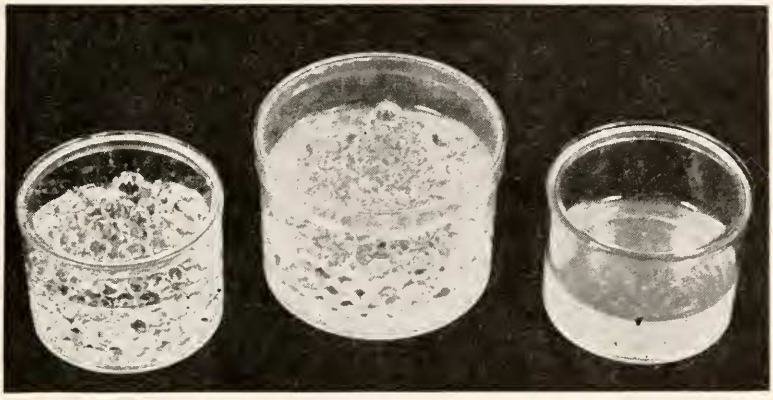

Fig. 1.

Rearing Containers.—Tabanid larvae are cannibalistic and must be reared in separate containers. Containers should be large enough to allow some freedom of movement. A rough rule of thumb used in the present studies was to select containers that gave a "crawl" distance equal to at least twice the length of the larva.

The circular containers used were made of crystal-clear rigid plastic, 2 in. in diameter and 1⅝ in. deep, with close-fitting snap-on lids.[3] Since the larvae tended to remain near the outer circumference, a "crawl" distance of about 6 in. was available, which was more than adequate even for larvae that were 50–60 mm long. Although larger containers were also used, no outstanding advantage was noted (Fig. 1).

These containers were filled to one-third to two-thirds of their volume with either agar or glass beads, the amount depending on the size of the larvae. Several $\frac{1}{16}$-in. holes drilled in the lid allowed for interchange of air. Although not necessary for

[3] Manufactured by Tri-State Plastic Molding Co., Henderson, Ky.

small larvae, air exchange was needed for larvae 25 mm or longer, especially when the containers were opened and examined only at two- to three-day intervals. These lids were not used for larvae under 10 mm long because the larvae were able to escape.

This technique has been used successfully for nearly 2 years in rearing larvae of *Tabanus lineola* Fabricius, *T. schwardti schwardti* Philip, *T. abdominalis* Fabricius, *T. proximus* Walker, and *Chlorotabanus crepuscularis* (Bequaert).

References Cited

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Nomenclature Notice

Possible use of plenary powers by the Commission is announced for: In Araneae—(1625) Suppression of *Drassus atropos* Walck. 1830. In Siphonaptera—(1618) Neotype for *Ceratophyllus soricis* Dale 1878; (1709) Type species for *Monopsyllus* Kolenati 1875; Suppression of *Ceratophyllus sciuri* Kol. 1856, *Monopsyllus sciuri* Kol. 1857, and *Ceratopsyllus monoctenus* Kol. 1856. In Lepidoptera—(1708) Suppression of *Papilio lintingensis* Osbeck 1765. In Coleoptera—(1720) Suppression of *Xyleborus* Bowdich 1865. In Diptera—(1706) Type-species for *Phasia* Latr. 1804; (1716) Type-species for *Chamaemyia* Meigen 1803. In Hymenoptera—(1710) Type-species for *Stizus* Latr. 1802–1803; (1711) *Id.* for *Diodontus* Curtis 1834; (1712) *Id.* for *Trichosis* Foerster 1868; (1713) *Id.* for *Prospaltella* Ashmead 1904.

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